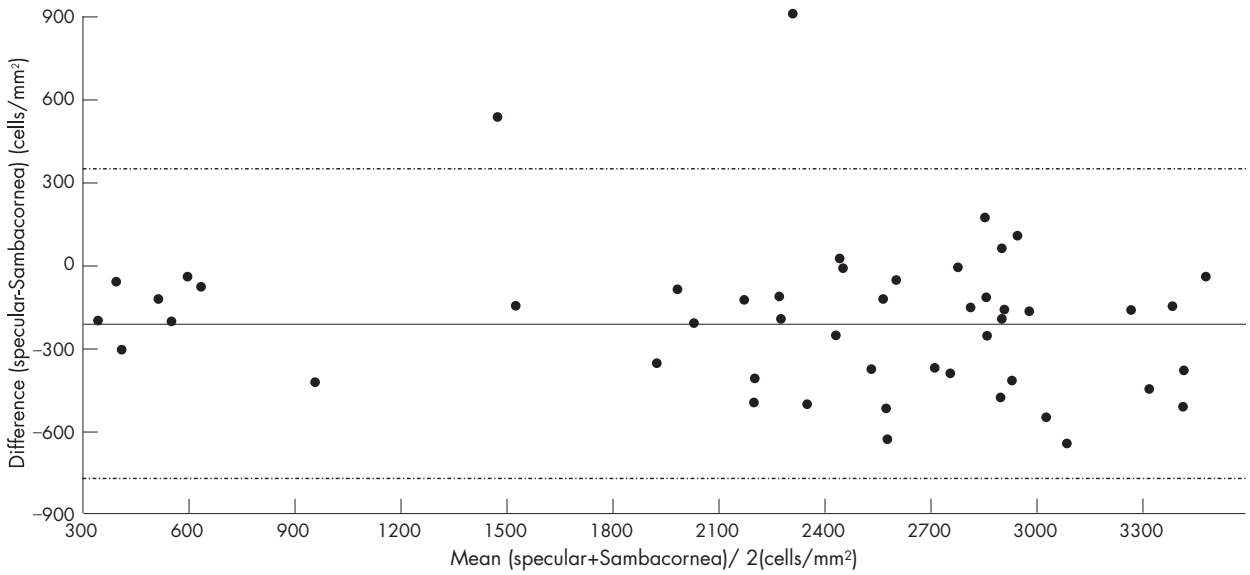

Figure 1 Difference against mean graph representing the agreement between cell counts of a Topcon specular microscope in vivo and the eyebank analyser Sambacornea ex vivo for the complete series of 51 corneas. The solid line represents the mean underestimation by specular microscopy compared with that by Sambacornea (221 cells/mm^2 95% CI 141 to 301). In all, 95% of the values of the difference between specular microscope and Sambacornea were contained between the two dashed lines (−779; +338 cells/mm^2) that represent the bounds of agreement between the two methods. This large interval between the two limits, even if partly due to the small sample size, highlighted the rather poor agreement between both methods.

Assessment of the human corneal endothelium: in vivo Topcon SP2000P specular microscope versus ex vivo sambacornea eye bank analyser

Comparison between assessment of donor tissue in eye banks and specular microscopy in the recipient is important to quantify the post-keratoplasty cell loss dynamics. Our aim was to determine the agreement between the in vivo non-contact specular microscope Topcon SP2000P and the computer-assisted eye bank endothelial analyser Sambacornea. We enrolled 51 future recipients of penetrating keratoplasty, and determined the endothelial cell density (ECD) and morphometry firstly in vivo with Topcon and then ex vivo with Sambacornea on the excised cornea stained with Alizarin Red. Specular microscopy was found to underestimate the ECD by 11%, (95% CI 6 to 15), whereas morphometric parameters did not differ.

Background

Endothelial cell loss after penetrating keratoplasty is commonly evaluated by non-contact specular microscopy. A comparison of tissue

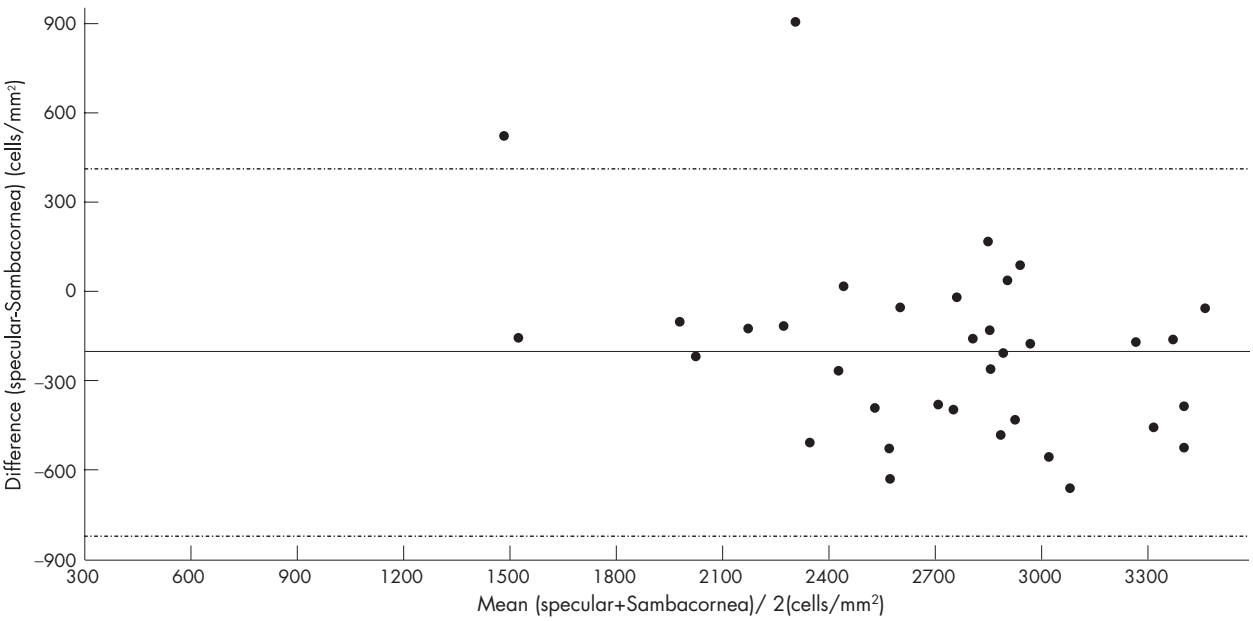

Figure 2 Difference against mean graph representing the agreement between both methods considering the 35 cases where the specular count was performed on ⩾75 cells, the minimum standard usually accepted for a reliable specular count.[6] The solid line represents the mean underestimation by specular microscopy compared with that by Sambacornea (202 cells/mm^2, 95% CI 94 to 310). The two bounds of agreement (−817; +413 cells/mm^2) as shown by the dashed lines are larger than that in figure 1 owing to the smaller sample size, but still showed a poor agreement between both methods.

assessment in eye banks and that by specular microscopy in the recipient is important to quantify the postoperative cell loss dynamics. In Europe, where organ culture is common, preoperative endothelial cell density (ECD) is assessed either by manual counting by observation through a microscope reticule or by using computer-assisted analysers.[1–4] We studied the agreement between ECD and morphometry determined by the non-contact small-field (0.08 mm^2) specular microscope Topcon SP2000P (Topcon, Tokyo, Japan) and that determined by a wide-field (1.2 mm^2/field) tri-image eye bank analyser Sambacornea (Sambatechnologies, Meylan, France) using light microscopy images.

Methods

Fifty one patients (comprising keratoconus 52%, lattice dystrophy 27% and others 21%) with somewhat clear central cornea were subjected to preoperative specular microscopy. The excised cornea was immediately (always within 30 min) stained with Alizarin Red and the central endothelium assessed by Sambacornea after appropriate calibration. For either analysis, an automated cell border detection was followed by manual touch-up of incorrectly drawn cells. Assessment of ECD, coefficient of variation (CV) of cell area and hexagonality was performed on the maximum number of cells possible. Agreement was determined using the Bland–Altman method.[5]

Results

Specular counts were performed on a mean (SD, range) of 102 (58, 4–223) cells compared with 2340 (1571, 163–6600) cells for Sambacornea ($p<0.001$). Specular microscopy underestimated the ECD by a mean of 221 cells/mm^2 (95% confidence interval (CI) 141 to 301) corresponding to an 11% (95% CI 6 to 15) underestimation independently of the ECD itself (fig 1). No correlation was found between the number of cells used for ECD determination with specular microscopy and the difference observed between the two methods ($r = -0.053$ and $p = 0.713$). In the subgroup ($n = 35$) of specular counts performed on ⩾75 cells (''minimum standard'' for reliability[6]), the mean underestimation was 202 cells/mm^2 (95% CI 94 to 310) corresponding to 6% (95% CI 1 to 11) (fig 2). The percentage of hexagonal cells and coefficent of variation were comparable: 55 (21, 0 to 100) and 32 (9, 10 to 56) for specular and 55 (11, 29 to 76) and 34 (13, 23 to 78) for Sambacornea ($p = 0.596$ and 0.588, respectively).

Comment

Few studies have compared specular and light microscopic endothelial assessments until now. A direct correlation has been shown between ECD from histological cross-sections and in vitro specular microscopy on eye bank corneas.[7] In the only morphometric comparison performed using wide-field contact specular microscopy, the authors did not find any significant difference between specular and post-staining cellular morphology, and in particular no evidence of tissue retraction.[8] Our protocol was designed to remove any possibility of tissue retraction as counting immediately followed excision. Calibration problems have been shown in Topcon SP2000P, producing underestimation in ECD values up to 9%, however, without any effect on morphometry.[9] Image quality has also been shown to influence morphometric parameters.[10] Using Sambacornea, counting of Alizarin-stained well-demarcated cells on a field 15 times larger than specular microscopy is expected to be more consistent with reality. Our morphometric findings remained comparable. The ECD underestimation by specular microscopy could help to explain a part of the step between the preoperative measurement in the eye bank and the first ECD measured postoperatively.

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BOOK REVIEW

Basic and Clinical Science Course: complete set

American Academy of Opthalmology. American Academy of Ophthalmology, £595 (soft cover). ISBN 1-56055-499-1

The American Academy series has always received a presence in most ophthalmology departmental libraries. Facilitating a further edition is a major undertaking, and the Academy has generated a more formative series of 13 volumes, providing clear objectives and study questions as good adjuncts to each volume. Although this series is written for residents in the USA, its value could be added if it appeals to trained ophthalmologists, allied professionals and, of course, the rest of the world. To gain such appeal, both content and style throughout so many volumes and disparate subjects (even within our small field) has to be uniform, and is a challenging task. So, on reading, I was impressed with the breadth covered. I enjoyed subjects that were not my subspecialty and was pleased to see that the style was reasonably consistent throughout. The series was well and appropriately illustrated. It was great to see a general medicine section covering aspects that have immense effects on both ophthalmic conditions and, of course, delivery of ophthalmic care.

The former section 13 of the series is now International Ophthalmology; I think it would be better as section 14 to maintain the emphasis of this important topic and to recognise the global nature of ophthalmology in the 21st century. It is a volume all residents should read.

To be critical, I did not believe that, in parts, subjects were evaluated using strict evidence-based criteria; the resident is led to key references only. On the other hand, perhaps this is a good aspect and makes the resident evaluate by further reading. The messages of therapeutic paradigms are mainly US market-based, and consequently, could have less effect globally. Further, the basic science volume was limited in the scope of increasing a resident's knowledge in certain chapters, which was disappointing.

However, I am sure most departments will add the improved and updated series to their shelves.

A Dick

NOTICES

Glaucoma

The latest issue of *Community Eye Health* (No 59) discussing new treatments for glaucoma in the developing world, with an editorial by leading specialist Richard Wormald. For further information please contact: Journal of Community Eye Health, International Resource Centre, International Centre for Eye Health, Department of Infectious and Tropical Diseases, London School of Hygiene and Tropical Medicine, Keppel Street, London WC1E 7HT, UK (tel: +44 (0)20 7612 7964; email: Anita.Shah@lshtm.ac.uk; online edition: www.jceh.co.uk). Annual subscription